

Appl. No.: 10/783,061
Amdt. dated June 25, 2007
Reply to Office Action of January 25, 2007

Amendments to the Specification:

Please replace paragraph [00025] with the following replacement paragraph:

[00025] Fig. 18 shows particle size distribution for mannitol particles obtained by spray-freezing through an Aeauspray™ ACCUSPRAY™ nozzle and drying by lyophilization. The particle size distribution was measured by laser diffraction.

Please replace paragraph [00063] with the following replacement paragraph:

[00063] Liquid formulations of the invention can be atomized by any of a variety of conventional procedures. For example, the liquid can be sprayed through a two-fluid nozzle, a pressure nozzle, or a spinning disc, or atomized with an ultrasonic nebulizer or a vibrating orifice aerosol generator (VOAG). In one embodiment, a liquid formulation is atomized with a pressure nozzle such as a BD Aeauspray™ ACCUSPRAY™ nozzle.

Please replace paragraph [000115] with the following replacement paragraph:

[000115] Dry powder formulations of whole, inactivated influenza virus A/PR/9/34 H1N1 particles were prepared in a spray-freeze dried batch process. A flu virus preparation was mixed into an aqueous solution, then atomized with a BD Aeauspray™ ACCUSPRAY™ nozzle. Liquid particle size data were obtained with a Sympatech diffractometer measuring at approximately 2 inches from the nozzle tip. The median diameter of particles produced at these concentrations was approximately 50 microns. A typical particle size distribution produced by the BD Aeauspray™ ACCUSPRAY™ nozzle is shown in Fig. 4. Liquid nitrogen was placed in a Virtis freeze-drying flask and the flask was positioned beneath the spray nozzle. The distance between the nozzle and liquid nitrogen was about three inches. The nebulized liquid droplets froze instantaneously upon contact with the liquid nitrogen. The flask was attached to a lyophilizer and immediately the excess liquid nitrogen was evaporated off. The frozen aerosols were typically dried within 48 hours and reached a moisture level below about 1 wt%.

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Please replace paragraph [000158] with the following replacement paragraph:

[000158] Aerodynamically Light Powder (ALP) formulations of rSEB vaccine were prepared by the method described above in Example 2. Briefly, a solution of the protein in a PBS sucrose mixture was nebulized using an ~~Aeauspray™~~ ACCUSPRAY™ nozzle, into a liquid nitrogen bath. The frozen particles thus formed were then lyophilized in a Vertis lyophilizer to remove moisture. Samples prepared by this method are referred to as spray-freeze-dried (SFD).

Please replace paragraph [000160] with the following replacement paragraph:

[000160] **SFD Procedure:**

1. Dissolve 200 mg of sucrose in 2 mL of DI water.
2. Thaw two vials of SEB (approximately 2 mL total volume).
3. Mix SEB with sucrose solution in a 5-ml ~~Aeauspray™~~ ACCUSPRAY™ syringe.
4. Mixed solution is sprayed into liquid N₂ according to one of the methods described herein.
5. Cool all necessary materials (spatula, scintillation vial, lyophilizer container, etc.) in dry ice to minimize heat transfer when collecting particles.
6. Collect frozen SFD particles and place in a scintillation vial.
7. Place scintillation vial in lyophilization chamber and attach chamber to a manifold lyophilizer. Dry ice should be placed around the lyophilizer container to ensure that the SFD particles stay frozen throughout primary drying.

Please replace paragraph [000180] with the following replacement paragraph:

[000180] The experiments were conducted as follows:

Materials:

Liquid “as received” rSEB vaccine (non-powder-processed) was obtained from USAMRIID, at 10 mg/ml in phosphate buffered saline (see above potency protocol).

Lyophilized rSEB vaccine was prepared by mixing the “as received” vaccine with pharmaceutical grade sucrose powder at a ratio of one part rSEB to 10 parts sucrose (w/w) to

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obtain a 10 mg/ml solution of rSEB with sucrose. The solution was lyophilized in a bench dryer for 3 days and milled as described above. ALP rSEB vaccine was produced by preparing a solution of rSEB with sucrose as described above, then spray-freeze-drying the solution using an Aeeuspray™ ACCUSPRAY™ system.